

IMCB's SARS-CoV RT-PCR Amplification Sites

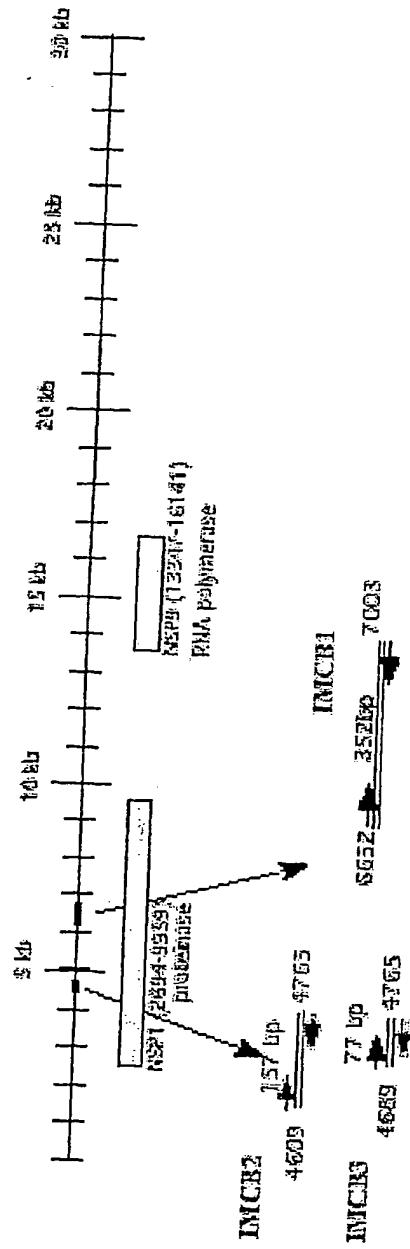


Figure 1

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GCCGTAGTGT CAGTATCATG ACCAGATGCT GTTACTACAT ATAATGGATA 50
CGGCATCACA GTCATAGTAG TGGTCTACGA CAATGATGTA TATTACCTAT
IMCB 3 Upper Primer

CCTCACTTCG TCATCAAGA CATCTGAGGA GCACCTTGTA GAAACAGTT 100
GGAGTGAAGC AGTAGTTCT GTAGACTCCT CGTGAAACAT CTTTGTCAAA
IMCB 3 TaqMan Probe sequence

CTTTGGCTGG CTCCTACGAG GATGGTCCT ATTCAAGGACA GCGTACAGAG 150
GAAACCGAGG GTCCTGCTT GATCCAGGA TAACTCCTGT CGCAGTGTCTC
template sequence IMCB 3 Lower Primer

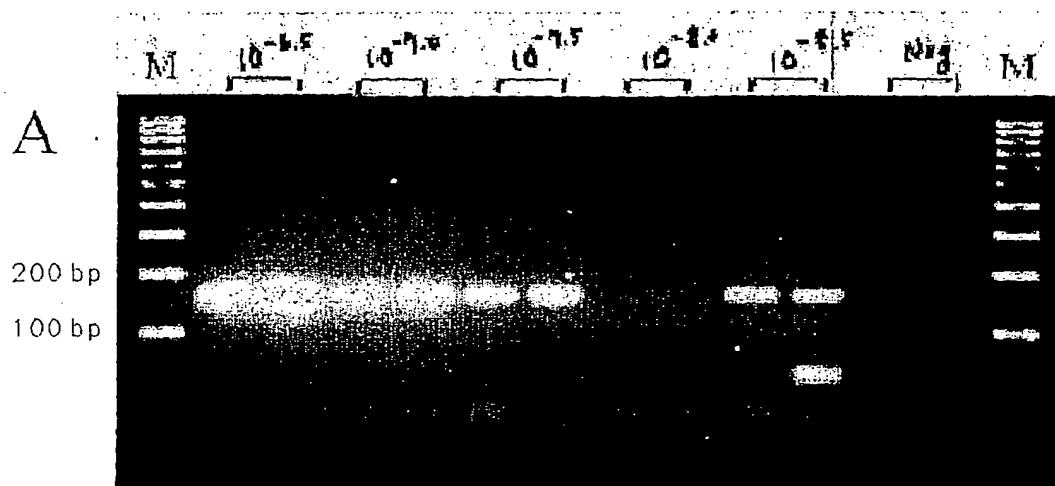
TAGGTG 157
CATCCAG
Template

Figure 2

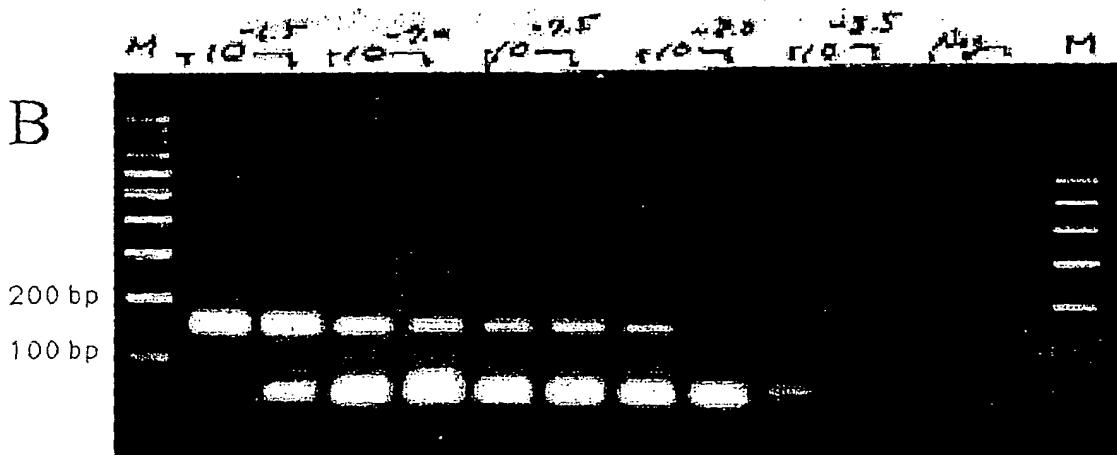
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SARS-CoV One-Step RT-PCR with IMCB 2 Primer Pair



Sensitivity12-4-27.jpg, 42 cycles



Sensitivity16-2-191.jpg, 42 cycles

rxn=5 ul, copies measured by ARTUS (and theoretical value)*

10(-6.0)=26.1 copies/rxn

10(-6.5)=12.6 copies/rxn, (8.2 copies/rxn)

10(-7.0)=1.96 copies/rxn, (2.6 copies/rxn)

10(-7.5)= 2.0 copies/rxn, (0.8 copies/rxn)

10(-8.0)= 0.07 copies/rxn, (0.25 copies/rxn)

10(-8.5)= Not detected, (0.08 copies/rxn)

*10(-6.0) is serially diluted by 10(-0.5) step.

Figure 3

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WO 2005/0591

App No.: NEW Docket No.: 4249-0117PUS1
Inventor: Masafumi INOUE et al.
Title: DETECTION OF SEVERE ACUTE RESPIRATORY
SYNDROME BY THE POLYMERASE CHAIN REACTION
NEW SHEET

Sheet 4 of 5 T/SG2004/000416

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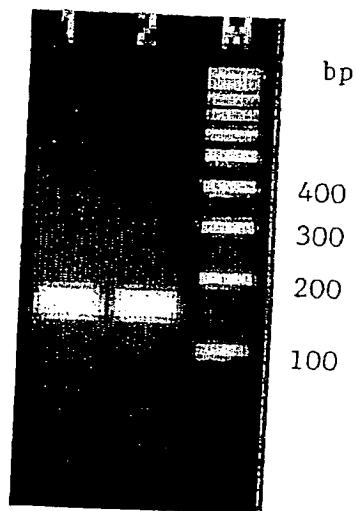
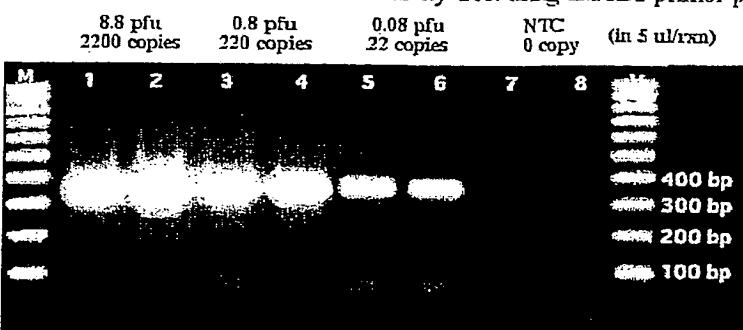


Figure 3C

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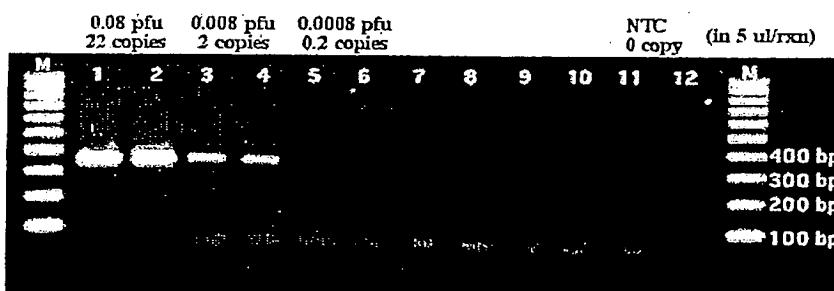
SARS CoV RT-PCR Detection Sensitivity Test using IMCB1 primer pair



L1-2: 8.8 pfu; L3-4: 0.8 pfu; L5-6: 0.08 pfu; L7-8: NTC in 5 µl sample. @1 pfu=250 copies

Fig. 4A

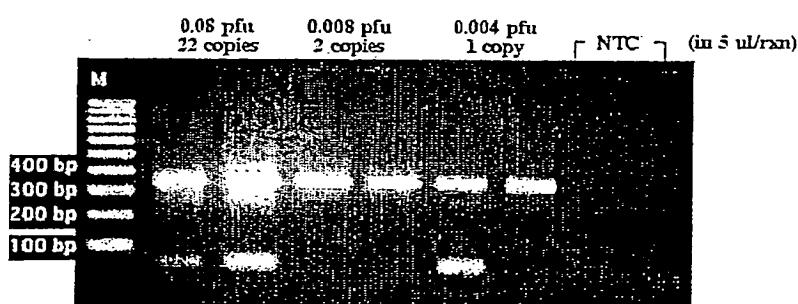
SARS CoV RT-PCR Detection Sensitivity Test using IMCB1 primer pair



L1-2: 0.08 pfu; L3-4: 0.008 pfu; L5-6: 0.0008 pfu; L7-8: 8.8 x 10⁴; L9-10: 8.8 x 10⁴; L11-12: NTC, in 5 µl sample. @1 pfu=250 copies

Fig. 4B

SARS CoV RT-PCR Detection Sensitivity Test using IMCB1 primer pair



L1-2: 0.08 pfu; L3-4: 0.008 pfu; L5-6: 0.0004 pfu; L7-8: NTC, in 5 µl sample. @1 pfu=250 copies

Fig. 4C

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